

Earliness and intensity of defoliation under the mild climate of Switzerland: a complete study on five cultivars over seven years

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Abstract. The objective of this work was to investigate the effects of earliness and intensity of defoliation on five *Vitis vinifera* cvs. – Pinot noir, Gamay, Merlot, Chasselas and Doral – under the mild-climate conditions of Switzerland. Between 2010 and 2016, intensive defoliation (removal of 6 basal leaves + 6 lateral shoots per shoot) was completed at three developmental stages of grapevine, i.e., pre-flowering, late flowering and bunch closure. Chasselas experiment also had a moderate pre-flowering defoliated treatment (removal of 3 basal leaves). In addition to the vintage effect, pre-flowering defoliation had tremendous consequences on the vine agronomic performance, mainly to the detriment of berry set: the yield was highly affected by the pre-flowering defoliation (approximately -30 % in comparison with no defoliation). The intensity of defoliation allowed the modulation of the impact on the yield. It also had a positive impact against millerandage, sunburn symptoms and Botrytis development. Berry skin thickness doubled and polyphenol concentration increased significantly. Due to pre-flowering defoliation, red wines were often preferred for their colour and structure in mouth. Meanwhile, this practice had negligible impact on white wine composition. In any case, pre-flowering defoliation did not have any negative impact on the wine parameters. In the context of this study, pre-flowering defoliation seems to be an interesting technique to reduce vigour and control high production potential. It also represents a prophylactic solution to reduce both chemical entrants and cluster-thinning costs.

1 Introduction

Grapevine defoliation in the cluster zone is usually realized between berry set and bunch closure to create an unfavourable microclimate for cryptogamic diseases, such as *Botrytis cinerea* and powdery mildew [1-3]. When completed after berry set, defoliation does not affect fruit set and yield [4-6]. However, grape growers are now interested in pre-flowering defoliation: this practice strongly affects berry set and berry number per bunch [7-10]. As a consequence, it limits the yield [11-13] and induces tremendous modifications in berry structure, i.e. skin thickness and skin-to-pulp ratio, and in berry composition (total soluble solids, acidity, and polyphenols) [12, 14-16]. Inducing strong competition for assimilates between vegetative and reproductive organs, pre-flowering defoliation also presents some risks: the major part of photosynthetically active foliage is removed at a time of high C and N requirements by the inflorescences, forcing the vine to further dig into its reserves in its wood and roots [16]. Consequently, during the year following defoliation, a lower vigour was noted

in some situations [12], as well as a lower bud fruitfulness [17, 18]. In other situations, no carryover effects could be observed because the vines had sufficient reserves [19].

Pre-flowering defoliation can drastically affect the must composition; the concentration of total soluble solids in the must usually increases in comparison to a non-defoliated control treatment, while acidity is decreased in some situations [12, 17, 20, 21]. Moreover, the accumulation of phenolic compounds increases [12, 22, 23], enhancing colour intensity and stability in red wines. Finally, the concentration of volatile compounds increases, possibly enhancing wine aroma quality [24]. However, the quantitative and qualitative parameters of the must and wine are not always affected in a significant manner [23, 25, 26].

Pre-flowering defoliation is a promising technique under the temperate conditions of Switzerland [16]. However, its impact on yield and grape composition seems to be unpredictable as a function of numerous biotic and abiotic factors, e.g., type of cultivar, climatic conditions, and period and intensity of defoliation [8, 27]. Considering the heterogeneity of the aforementioned

results and the risk of excessive yield loss resulting from this practice, the present work was required to investigate the effects of pre-flowering defoliation on a selection of five local Swiss cv. under local Swiss conditions, in comparison to alternative defoliation timing and intensity, with particular attention paid to its effects on yield reduction and must composition.

2 Material and methods

2.1 Vineyard site and experimental design

Five experiments was conducted between 2010 and 2016 in three experimental vineyards of Agroscope on five field-grown *Vitis vinifera* L. cv (Table 1). The vines were grafted onto rootstock 3309C, planted at a density of 5880 vines/ha (except for Merlot at 5200 vine/ha) and pruned using a single-Guyot training system (except for Pinot noir with Cordon Royat). The canopy was trimmed to 110 cm in height. The lateral shoots were removed from the fruiting zone during the berry-set stage (BBCH 71) as a normal practice in the region.

Table 1. Description of the fives experiments

Cultivar	Vineyard	Trial period	Plantation date
Chasselas	Pully	2011-16	2007
Doral	Changins	2011-15	2003
Pinot noir	Pully	2010-15	1991
Gamay	Changins	2010-16	2007
Merlot	Gudo	2011-16	2006

The experiment was structured as a randomized block design, including four blocks with four treatments of at least 10 vines each (A, B, C, D); a fifth treatment (E) was applied on Chasselas only (Table 2). Treatments consisted at removing leaves from the fruiting zone as follow.

Table 2. Description of the different treatments.

Variante	Defoliation timing	Defoliation intensity
A	Control non defoliated	-
B	Pre-flowering (BBCH 57)	Intensive, 6 leaves
C	Late Flowering (BBCH 67-69)	Intensive, 6 leaves
D	Bunch closure (BBCH 77)	Intensive, 6 leaves
E	Pre-flowering (BBCH 57)	Moderate, 3 leaves

2.2 Field measurement

All measurements were realized per repetition. The phenological stages flowering and veraison were dated. The bud fruitfulness was estimated (average number of clusters per shoot). The potential yield ($Yield_{estim}$) was

estimated in July (before bunch closure) from a sample of 50 berries and 10 bunches per replicate using the following formula:

$$Yield_{estim} = \frac{\frac{cluster\ wt_{July} * berry\ wt_{harv}}{berry\ wt_{July}} * cluster\ nb_{vine}}{plantation\ density * 1000} \quad (1)$$

Berry w_{July} and bunch w_{July} are the average berry and bunch weights in July (stage BBCH 75-77), respectively, and berry w_{harv} is the average berry weight at harvest for each cultivar since 2005. Cluster nb_{vine} is the cluster number per vine.

The chlorophyll index (N-tester, Yara, France), which permitted the monitoring of the chlorophyll concentration throughout the season, was measured once a month in the medial zone of the canopy. The light-exposed leaf area (m^2/m^2 of soil) was determined using Carbonneau's method [28]. The vigour of the vines was assessed during winter by weighing 10 one-meter long pruned canes and was expressed in grams per meter (g/m). A leaf diagnosis was carried out at veraison on 25 leaves (limb + petiole) per treatment from the medial part of the canopy and analysed at the registered laboratory Sol-Conseil (Gland, CH) in order to assess the N, P, K, Ca and Mg contents. When symptoms of millerandage, sunburn, or an attack by *Botrytis cinerea* occurred, it was quantified per replicate by the percentage of symptoms per cluster on 25 clusters. In 2013 and 2015, cluster samples were collected before harvest to evaluate berry skin thickness in the treatments A, B and D of Pinot noir and Chasselas. Three berries from three clusters per treatment were prepared according to Roland and Vian [29]. Semi-thin sections were observed using a light microscope (Leica DMLB, Leica Microsystems, Heerbrugg, Switzerland): four sites per berry were randomly measured from the upper epidermis to the limit between the hypodermis (tangential cell layer) and mesocarp (pulp cells).

At harvest, grape extract analyses were performed on Pinot noir, as detailed in Verdenal et al. [16], at the Agroscope laboratory to determine the following parameters: total polyphenolic content, glutathione, total free anthocyanins and anthocyanin profile. Standard must analyses were also performed using an infrared spectrophotometer (FOSS WinescanTM): berry weight, titratable acidity (TA eq. tartaric acid), tartaric and malic acids, total soluble solids (TSS), pH and yeast available nitrogen (YAN). Finally, approximately 60 kg of grapes from each treatment were harvested and vinified separately at the Agroscope winery, as detailed in Verdenal et al. [16]. Finished wines were analysed using an infrared spectrophotometer (FOSS WineScanTM) for the following parameters: alcohol, dry weight, pH, volatile acid, titratable acidity, tartaric, malic and lactic acids, glycerol, and free and combined SO_2 . Wine sensory analysis was realized by the Agroscope tasting panel.

The data description and the significance of the differences between treatments, sites and vintages were statistically evaluated using analysis of variance (ANOVA, p values < 0.05), multiple comparison Newman-Keuls test and principle component analysis (PCA) realised with ©XLSTAT 2015.1.02.

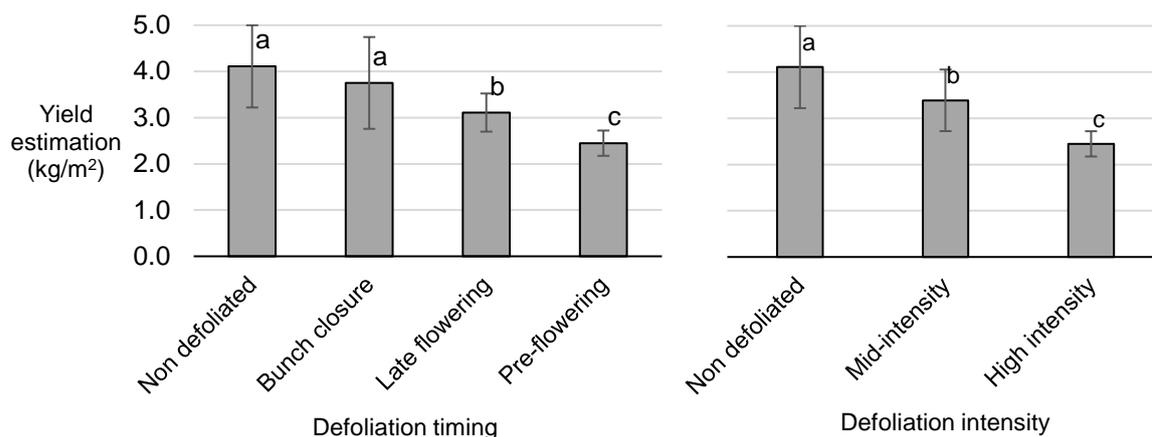


Figure 1. Impact of defoliation timing and intensity on yield potential of Chasselas (Pully), estimated before bunch thinning. 2013-2016 averages \pm SD. Treatments with different letters are significantly different (Newman-Keuls test, $P < 0.05$).

3 Results and discussion

3.1 Yield parameters

For all cultivars, pre-flowering defoliation had a significant impact on berry-set rate. In the example of Chasselas, treatment B presented different cluster structures in comparison to those of the other treatments (A, C, D, and E): clusters were globally smaller (-30 % wt.) and had fewer berries (-36 %), although their berries were always smaller (depending on the cultivar). As a consequence, the average 2013–2016 yield potential estimation showed a 40 % loss under the pre-flowering treatment (B) in comparison to that under the control treatment (A), a 24 % loss under the late-flowering treatment (C) and no significant loss under the bunch-closure treatment (D) (Figure 1). The mid-intensity treatment (E) modulated the impact of pre-flowering defoliation with only an 18 % loss.

3.2 Plant behaviour and carryover effects

Phenology was affected by the defoliation period in all cultivars. In the example of Pinot noir, pre-flowering treatment (B) consistently showed earliness: at flowering stage, 72 ± 8 % of flowering was completed against an average of 57 ± 13 % in the three others treatments (A, C, D). This tendency was confirmed at veraison stage; the two latest defoliation treatments (C) and (D) showed a delay (-9 % on average) in comparison to the pre-flowering and control treatments (B) and (A).

Carryover effects could be observed only in Chasselas trial: in that case, intensive pre-flowering defoliation (B) induced a slightly lower bud fruitfulness (-0.1 bunch/shoot in comparison to that of the other treatments). Despite the variability between vintages, defoliated treatments (B, C and D) also had a lower trimming weight (an average of 571 ± 205 g versus 682 ± 236 g under the non-defoliated treatment A). Mid-intensity defoliation (E) modulated the impact on the trimming weight (613 ± 214 g). Moreover, both the high-intensity and earliness of defoliation (B) induced lighter pruning weights during the winter (54 ± 7 g/m under treatment B versus 64 ± 7 g/m under treatment A). However, vine sustainability was not affected.

High millerandage rates were recorded in 2010 and 2013: both years, the earlier the defoliation, the lower the millerandage rate, while no differences were noticed between the control and the bunch-closure treatments (A and D) (Figure 2).

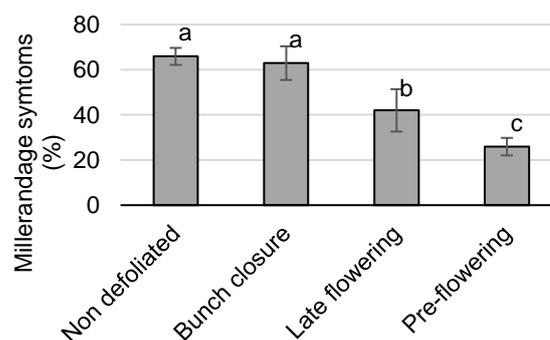


Figure 2. Impact of the defoliation period on the development of millerandage symptoms on Pinot noir (Pully). 2013 data \pm SD. Treatments with different letters are significantly different (Newman-Keuls test, P value < 0.05).

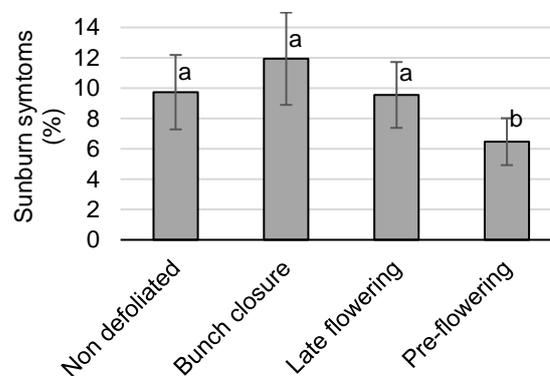


Figure 3. Impact of the defoliation period on the sunburn symptoms on Gamay (Changins). Average 2012, 2014 and 2016 \pm SD. Treatments with different letters are significantly different (Newman-Keuls test, P value < 0.05).

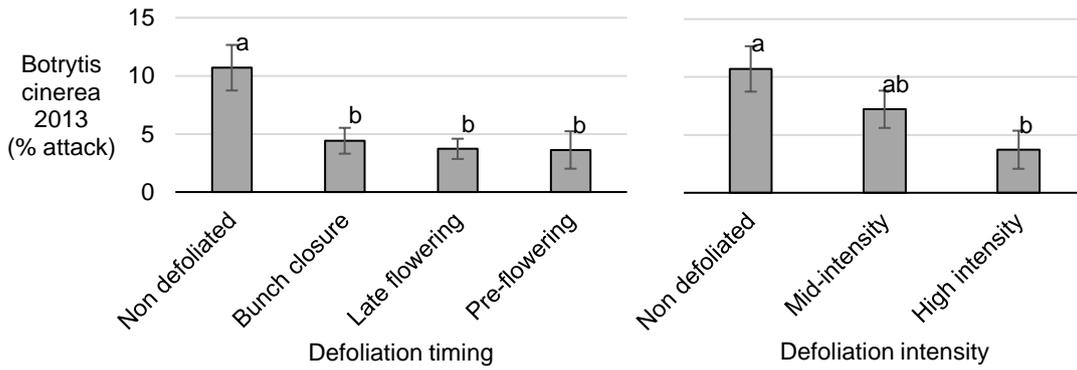


Figure 4. Impact of defoliation timing and intensity on Botrytis attack on the clusters of Chasselas (Pully). 2013 averages \pm SD. Treatments with different letters are significantly different (Newman-Keuls test, $P < 0.05$).

Higher rates of sunburn symptoms appeared on the grapes in 2012, 2014 and 2016: each year, significantly less symptoms could be observed in the pre-flowering treatment B (Figure 3).

The 2013 bunch rot attack on Chasselas confirmed the defoliation efficiency against Botrytis cinerea (Figure 4): the control treatment (A) had an 11 % loss due to grey mould, while the three defoliated treatments had a loss of less than 4 % loss. This resistance was clearly related to defoliation intensity, which reduces humidity and creates an unfavourable microclimate for fungus inoculation.

Defoliation treatments significantly affected berry skin thickness (P value < 0.0001), while the vintage effect was negligible. In the case of Pinot noir, berries in the control treatment (A) presented thinner skins (two-year average, $110 \pm 8 \mu\text{m}$), followed by the bunch-closure treatment (D) ($149 \pm 13 \mu\text{m}$) and then the pre-flowering treatment (B) ($219 \pm 17 \mu\text{m}$) (Figure 5). These results had consequences on grape extract chemical composition as presented below.

3.3 Must composition and wine tasting

Concerning the white cultivars (Chasselas and Doral), inconsistent and negligible differences could be observed in terms of must composition. In the white wines, the differences were insignificant and no wine was preferred to the others. Gamay musts and wines did not present any significant difference. On the other hand, the red cultivars Pinot noir and Merlot, the musts from the non-defoliated control were frequently more acidic. Total polyphenols (Folin index), particularly anthocyanins, were more concentrated in the wines from pre-flowering treatments (Figure 6), giving more appreciated wines in the end in

terms of color intensity, fruity, mouth feeling and overall hedonistic appreciation (Table 3).

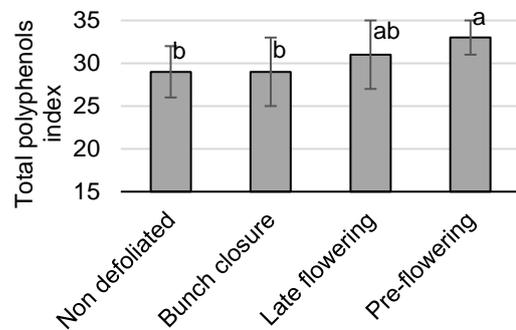


Figure 6. Impact of defoliation timing on the polyphenol concentration in the wines of Pinot noir (Pully). 2013-2015 averages \pm SD. Treatments with different letters are significantly different (Newman-Keuls test, $P < 0.05$).

Table 3. Main distinctive criteria from wine sensory analysis of Pinot noir (Pully). Quotes between 1 and 7; 6-year average. Numbers in the same column with different letters are significantly different (Newman-Keuls test, $P < 0.05$).

	Color intensity	Tanins structure	Hedonistic impression
Non defoliated	4.1 c	3.1 b	4.0 b
Bunch closure	4.2 bc	3.1 b	4.2 ab
Late flowering	4.3 ab	3.4 a	4.2 ab
Pre-flowering	4.4 a	3.4 a	4.3 a

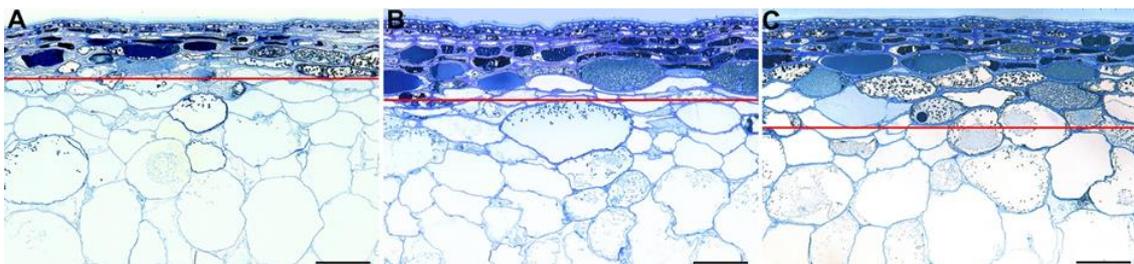


Figure 5. Semi-thin sections of berry epidermal cells showing the effects of two defoliation stages on berry skin thickness on Pinot noir at harvest 2013. A: non-defoliated control (treatment A); B: bunch-closure defoliation (treatment D); C: pre-flowering defoliation (treatment B). Scale bars represent 100 μm .

4 Discussion

Besides the cultivar effect, the variability of the climate modulates the floral initiation (year n-1) and the rate of berry set (year n), which both determine the initial yield potential. Intensive pre-flowering defoliation usually led to an approximately 30 % yield loss in comparison to the non-defoliated control, whichever cultivar and independently from the initial yield potential. In other words, the yield loss was proportional to the potential of production. The intensity of pre-flowering defoliation allowed for the modulation of its impact and can prevent an excessive yield loss. These results are possibly related to the competition between the growing canopy and the inflorescences for assimilates during the early season. As a consequence, this practice should not be recommended on too young or not enough vigorous vines.

Pre-floral defoliation reduced acidity and increased polyphenolic concentration in red wines, as mentioned in the literature [14, 15]. However, concerning white cultivars, their berry skin contain no anthocyanins, and there is usually no skin maceration during the winemaking. These two points greatly reduce the role of pre-flowering defoliation on wine quality, as there is no oenological interest in terms of polyphenol accumulation and colour intensity in white wine, in contrast to red wine. As a confirmation in the present trial, no difference was observed between the wines, neither for Doral nor for Chasselas.

5 Conclusion

Despite the variability of its impact – mainly due to the climate unpredictability and the cultivar – pre-flowering defoliation resulted in tremendous effects on vine physiology. It represents an interesting sustainable practice to control yield and enhance resistance to pathogens under the temperate climate of Switzerland. It also had a positive impact on the sensory profile of the red wines (higher colour intensity, lower acidity). The intensity of pre-flowering defoliation is a good leverage to prevent an excessive yield loss. However, this practice also presents a part of risks, as it can affect vine vigour and thus can potentially reduce vine sustainability under restrictive conditions. However, pre-flowering defoliation never had a negative impact on the must and wine composition in the context of these experiments.

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References

1. B. W. Zoecklein, T. K. Wolf, N. W. Duncan, J. M. Judge, M. K. Cook, *Am. J. Enol. Vitic.* **43** (1992)
2. D.C. Percival, J.A. Sullivan, K.H. Fisher, *Vitis* **32** (1993)
3. M. Sternad Lemut, P. Sivilotti, L. Butinar, J. Laganis, U. Vrhovsek *Aust. J. Grape Wine Res.* **21** (2015)
<https://doi.org/10.1111/ajgw.12148>
4. H. Feng, F. Yuan, P.A. Skinkis, M.C. Qian, *Food Chem.* **173** (2015)
<https://doi.org/10.1016/j.foodchem.2014.09.149>
5. E. Nicolosi, A. Continella, A. Gentile, A. Cicala, F. Ferlito, *Sci. Hort.* **146** (2012)
<https://doi.org/10.1016/j.scienta.2012.07.033>
6. J. Tardaguila, M.P. Diago, F. Martinez de Toada, S. Poni, M. Vilanova, *J. Int. Sci. Vigne Vin* **42** (2008)
<https://doi.org/10.20870/oeno-one.2008.42.4.810>
7. I. Gómez, J. Revert, M.D. Esteve, M.D. Climent, A. Martínez, J. Jiménez, D.S. Intrigliolo, *Acta Hort.* **931** (2012)
<https://doi.org/10.17660/ActaHortic.2012.931.42>
8. Y. Kotseridis, A. Georgiadou, P. Tikos, S. Kallithraka, S. Koundouras, *J. Agric. Food Chem.* **60** (2012)
<https://doi.org/10.1021/jf300605j>
9. S. Poni, F. Bernizzoni, *J. Int. Sci. Vigne Vin* **44** (2010)
<https://doi.org/10.20870/oeno-one.2010.44.1.1458>
10. P. Sabbatini, G.S. Howell, *HortScience* **45** (2010)
11. S. Poni, L. Casalini, F. Bernizzoni, S. Civardi, C. Intriери, *Am. J. Enol. Vitic.* **57** (2006)
12. A. Palliotti, T. Gardi, J.G. Berrios, S. Civardi, S. Poni, *Sci. Hort.* **145** (2012)
<https://doi.org/10.1016/j.scienta.2012.07.019>
13. B. Basile, G. Caccavello, M. Giaccone, M. Forlani, *Am. J. Enol. Vitic.* **66** (2015)
14. K. Šuklje, G. Antalick, Z. Coetzee, L.M. Schmidtke, H. Baša Česnik, J. Brandt, W. J. du Toit, K. Lisjak, A. Deloire, *Austr. J. Grape Wine Res.* **20** (2014)
15. B. L. Komm, M. M. Moyer, *Am. J. Enol. Vitic.* **66** (2015)
16. T. Verdenal, V. Zufferey, A. Dienes-Nagy, K. Gindro, S. Belcher, F. Lorenzini, J. Rösti, C. Koestel, J.-L. Spring, O. Viret, *OENO One* **51** (2017)
17. D. Risco, D. Pérez, A. Yeves, J.R. Castel, D.S. Intrigliolo, *Aust. J. Grape Wine Res.* **20** (2014)
<https://doi.org/10.1111/ajgw.12049>
18. D. Uriarte, J. Picón, L.A. Mancha, J. Blanco, M.H. Prieto, D. Moreno, E. Gamero, E. Valdés, D. Risco, J.R. Castel, D.S. Intrigliolo, *Acta Hort.* **931** (2012)
<https://doi.org/10.17660/ActaHortic.2012.931.33>
19. D. Acimovic, L. Tozzini, A. Green, P. Sivilotti, P. Sabbatini, *Aust. J. Grape Wine Res.* **22** (2016)
<https://doi.org/10.1111/ajgw.12235>
20. B. Bravetti, V. Lanari, E. Manni, O. Silvestroni, *Acta Hort.* **931** (2012)
<https://doi.org/10.17660/ActaHortic.2012.931.37>
21. M.P. Diago, M. Vilanova, J. Tardaguila, *Am. J. Enol. Vitic.* **61** (2010)
22. M. Sternad Lemut, K. Trost, P. Sivilotti, P. Arapitsas, U. Vrhovsek, *J. Sci. Food Agric.* **93** (2013)

23. M.I. Talaverano, D. Moreno, F.J. Rodríguez-Pulido, M.E. Valdés, E. Gamero, M.J. Jara-Palacios, F.J. Heredia, *Sci. Hortic.* **209** (2016)
<https://doi.org/10.1016/j.scienta.2016.06.013>
24. M. Vilanova, M.P. Diago, Z. Genisheva, J.M. Oliveira, J. Tardaguila, *J. Sci. Food Agric.* **92** (2012)
<https://doi.org/10.1002/jsfa.4673>
25. D. Moreno, M. Vilanova, E. Gamero, D.S. Intrigliolo, M.I. Talaverano, D. Uriarte, M.E. Valdes, *Am. J. Enol. Vitic.* **66** (2015)
<https://doi.org/10.5344/ajev.2014.14087>
26. P. Sivilotti, J.C. Herrera, K. Lisjak, H.B. Cesnik, P. Sabbatini, E. Peterlunger, S.D. Castellarin, *J. Agric. Food Chem.* **64** (2016)
<https://doi.org/10.1021/acs.jafc.6b01013>
27. B. Hed, H.K. Ngugi, J.W. Travis, *Am. J. Enol. Vitic.* **66** (2015)
<https://doi.org/10.5344/ajev.2014.14034>
28. A. Carbonneau, *Prog. Agric. Vitic.* **112** (1995)
29. J. Roland, B. Vian, In: *Electron Microscopy of Plant Cells*. (Academic Press, London, 1991)
<https://doi.org/10.1016/B978-0-12-318880-9.50006-5>